

Version with Markings to Show Changes Made:

In the Specification

Please amend page 1, lines 3-4 of the specification as follows:

This application is a divisional of U.S. Application Serial No. 09/193,612, filed on November 17, 1998, which claims the benefit of U.S. Provisional Application Serial No. 60/098,235 filed August 28, 1998 and U.S. Provisional Application Serial No. 60/065,628 filed November 18, 1997, all of which [is] are herein incorporated by reference.

Please amend page 4, lines 13-17 of the specification as follows:

Figure 1 provides a chimeric oligonucleotide comprising three intervening blocks of RNA residues for modification of the maize EPSPS gene to a herbicide resistant form of the gene by converting two nucleotides at each of two amino acid residues of the target sequence. The linear (SED ID NO: 1) and active forms of the oligonucleotide are provided. A fragment of the maize EPSPS amino acid sequence is translated beneath the active oligonucleotide and is set forth in SEQ ID NO: 22.

Please amend page 4, lines 18-22 of the specification as follows:

Figure 2 provides a chimeric oligonucleotide comprising three intervening blocks of RNA residues for modification of the maize EPSPS gene to a herbicide resistant form of the gene by converting one nucleotide at each of two amino acid residues of the target sequence. The linear (SEQ ID NO: 2) and active forms of the oligonucleotide are provided. A fragment of the maize EPSPS amino acid sequence is translated beneath the active oligonucleotide and is set forth in SEQ ID NO: 23.

Please amend page 4, lines 23-27 of the specification as follows:

Figure 3 provides a chimeric oligonucleotide comprising two intervening blocks of RNA residues for modification of the maize EPSPS gene to a herbicide resistant form of the gene by

converting two nucleotides at each of two amino acid residues of the target sequence. The linear (SEQ ID NO: 3) and active forms of the oligonucleotide are provided. A fragment of the maize EPSPS amino acid sequence is translated beneath the active oligonucleotide and is set forth in SEQ ID NO: 23.

Please amend page 4, lines 28-32 of the specification as follows:

Figure 4 provides a chimeric oligonucleotide comprising two intervening blocks of RNA residues for modification of the maize EPSPS gene to a herbicide resistant form of the gene by converting one nucleotide at each of two amino acid residues of the target sequence. The linear (SEQ ID NO: 4) and active forms of the oligonucleotide are provided. A fragment of the maize EPSPS amino acid sequence is translated beneath the active oligonucleotide and is set forth in SEQ ID NO: 23.

Please amend pages 4 and 5, lines 33-34 and 1-4, respectively, of the specification as follows:

Figure 5 provides a chimeric oligonucleotide comprising two intervening blocks of RNA residues for modification of the maize EPSPS gene to a herbicide resistant form of the gene by converting two nucleotides at each of two amino acid residues of the target sequence. The first amino acid residue target is within a DNA region while the second amino acid residue target is within an RNA region. The linear (SEQ ID NO: 5) and active forms of the oligonucleotide are provided. A fragment of the maize EPSPS amino acid sequence is translated beneath the active oligonucleotide and is set forth in SEQ ID NO: 22.

Please amend page 5, lines 5-10 of the specification as follows:

Figure 6 provides a chimeric oligonucleotide comprising two intervening blocks of RNA residues for modification of the maize EPSPS gene to a herbicide resistant form of the gene by converting one nucleotide at each of two amino acid residues of the target sequence. The first amino acid residue target is within a DNA region while the second amino acid residue target is

within an RNA region. The linear (SEQ ID NO: 6) and active forms of the oligonucleotide are provided. A fragment of the maize EPSPS amino acid sequence is translated beneath the active oligonucleotide and is set forth in SEQ ID NO: 24.

Please amend page 5, lines 11-16 of the specification as follows:

Figure 7 provides a chimeric oligonucleotide comprising two intervening blocks of RNA residues for modification of the maize EPSPS gene to a herbicide resistant form of the gene by converting two nucleotides at each of two amino acid residues of the target sequence. The first amino acid residue target is within an RNA region while the second amino acid residue target is within a DNA region. The linear (SEQ ID NO: 7) and active forms of the oligonucleotide are provided. A fragment of the maize EPSPS amino acid sequence is translated beneath the active oligonucleotide and is set forth in SEQ ID NO: 22.

Please amend page 5, lines 17-22 of the specification as follows:

Figure 8 provides a chimeric oligonucleotide comprising two intervening blocks of RNA residues for modification of the maize EPSPS gene to a herbicide resistant form of the gene by converting one nucleotide at each of two amino acid residues of the target sequence. The first amino acid residue target is within an RNA region while the second amino acid residue target is within a DNA region. The linear (SEQ ID NO: 8) and active forms of the oligonucleotide are provided. A fragment of the maize EPSPS amino acid sequence is translated beneath the active oligonucleotide and is set forth in SEQ ID NO: 24.

Please amend page 5, lines 23-27 of the specification as follows:

Figure 9 provides a chimeric oligonucleotide for a single amino acid modification of the maize EPSPS gene to a herbicide resistant form of the gene. The amino acid target here corresponds to the first of the two amino acid residues targeted by the chimeric oligonucleotides in Figures 1-8. The linear (SEQ ID NO: 9) and active forms of the oligonucleotide are provided.

A fragment of the maize EPSPS amino acid sequence is translated beneath the active oligonucleotide and is set forth in SEQ ID NO: 25.

Please amend page 5, lines 28-32 of the specification as follows:

Figure 10 provides a chimeric oligonucleotide for a single amino acid modification of the maize EPSPS gene to a herbicide resistant form of the gene. The amino acid target here corresponds to the second of the two amino acid residues targeted by the chimeric oligonucleotides in Figures 1-8. The linear (SEQ ID NO: 10) and active forms of the oligonucleotide are provided. A fragment of the maize EPSPS amino acid sequence is translated beneath the active oligonucleotide and is set forth in SEQ ID NO: 26.

Please amend pages 5 and 6, lines 33-34 and 1-2, respectively, of the specification as follows:

Figure 11 provides a chimeric oligonucleotide for a single amino acid modification at amino acid position 621 of the maize AHAS gene to a herbicide resistant form of the gene. The linear (SEQ ID NO: 11) and active forms of the oligonucleotide are provided. A fragment of the maize AHAS amino acid sequence is translated beneath the active oligonucleotide and is set forth in SEQ ID NO: 27.

Please amend page 6, lines 3-6 of the specification as follows:

Figure 12 provides a chimeric oligonucleotide for a single amino acid modification at amino acid position 165 of the maize AHAS gene to a herbicide resistant form of the gene. The linear (SEQ ID NO: 12) and active forms of the oligonucleotide are provided. A fragment of the maize AHAS amino acid sequence is translated beneath the active oligonucleotide and is set forth in SEQ ID NO: 28.

Please amend page 6, lines 7-10 of the specification as follows:

Figure 13 provides a chimeric oligonucleotide for a single nucleotide modification which converts a stop codon to a codon encoding tyrosine in a transgene target previously introduced into maize (see text). The linear (SEQ ID NO: 13) and active forms of the oligonucleotide are provided. A fragment of the maize AHAS amino acid sequence is translated beneath the active oligonucleotide and is set forth in SEQ ID NO: 29.

Please amend page 17, lines 8-24 of the specification as follows:

For the transgene target a translational fusion between phosphinothricin-N-acetyltransferase, *pat* (Wohleben *et al.* (1988) *Gene* 70:25-37) and the green fluorescence protein, GFP (Prasher *et al.* (1992) *Gene* 111:229-233) was created. *pat* is a functional analog of the *bar* gene that similarly detoxifies Bialaphos. The coding sequences of GFP and *pat* have been modified to utilize maize preferred codons to enhance expression, these modified genes are referred to as GFPm and mo-PAT respectively. See, for example, U.S. application 09/003,287. A fusion was initially created by cloning the 3' BglII site in mo-PAT to a 5' flanking BamHI site on GFPm. Site directed mutagenesis (MORPH kit, 5'-3' Boulder, CO) was then used to remove the start codon (ATG) from GFPm to ensure low background expression of GFPm in target lines. Using the oligonucleotide PHN14593, 5'CGGTGACGCAGATCTATCCAACATTGTCCAAGGGC3' (SEQ ID NO: 14), the BglII site was recreated in mo-PAT and the start codon of GFPm was removed simultaneously. Four amino acids, YPTS, form the junction in the mo-PAT/GFPm sequence. The vector pPHP10699, is a positive control mo-PAT/GFPm fusion cloned under the control of the maize Ubiquitin-1 promoter and pinII terminator in a pUC-derived plasmid backbone.

Please amend pages 17 and 18, lines 25-34 and 1-2, respectively, of the specification as follows:

To create the target sequence for correction, the native *pat* stop codon (TGA) was inserted in the junction of mo-PAT/GFPm. Site-directed mutagenesis of pPHP10699 with

oligonucleotide PHN16214, 5'GGTGACGCAGATCTAGGTACCATCGTCCAAGGGCGAG3' (SEQ ID NO: 15), was used to change the junction sequence from YPTS to *VPS and to introduce a *KpnI* site adjacent to the stop codon. This creates a sequence that only expresses mo-PAT, but with correction to remove the stop codon, GFPm expression results. When making corrections to this target, changing the TAG stop codon to TAC also knocks out the *KpnI* site and creates a novel *SnaBI* site. The vector pPHP11207, contains the mo-PAT/TAG/GFPm target sequence, cloned with the maize Ubiquitin-1 promoter and pinII terminator, and inserted into a superbinary vector pSB1 for *Agrobacterium* mediated transformation of maize (Ishida *et al.* (1996) *Nature Biotech.* 14:745-750).

Please amend page 20, lines 14-27 of the specification as follows:

PCR amplification and sequence analysis - Target sequences were amplified from the extracted genomic DNA of putative events, by *Pwo* or *Taq* polymerase (Boehringer Mannheim, Indianapolis, IN) with 30 cycles of 35 seconds at 95°C, 35 seconds at 60°C, and 35 seconds at 72°C using a MJ thermocycler (MJ Research, Watertown, MA). For the AHAS621 target, primers common to both AHAS108 and AHAS109 were designed with the following sequences: 5'GCAGTGGGACAGGTTCTAT (PHN21971) (SEQ ID NO: 16) and 5'AGTCCTGCCATCACCATCCA (PHN21972) (SEQ ID NO: 17). For the AHAS165 target, the following primers were used: 5'ACCCGCTCCCCCGTCAT (PHN21973) (SEQ ID NO: 18) and 5'ATCTGCTGCTGGATGTCCTTGG (PHN21974) (SEQ ID NO: 19). For the moPAT/GFPm target, primers used were: 5'CGCAACGCCTACGACTGGA (PHN21976) (SEQ ID NO: 20) and 5'TGATGCCGTTCTTCTGCTTGTC (PHN21978) (SEQ ID NO: 21). PCR fragments were purified and either cloned (see below) or directly sequenced in both directions on an ABI 377 automated sequencer.

In the Claims

2. (Amended) The chimeric oligonucleotide of claim [1] 6, wherein said [molecule] chimeric oligonucleotide comprises at least one region of contiguous unpaired bases disposed so that the unpaired region separates the chimeric oligonucleotide into a first and a second strand.

3. (Amended) The chimeric oligonucleotide of claim 2, wherein said first and said second blocks of RNA [blocks] residues are comprised of a 2'-O or 2'-OMe ribose.

4. (Amended) The chimeric oligonucleotide of claim 2, wherein said first and said second blocks of RNA [block] residues [comprises] comprise at least 5 contiguous nucleotides.

5. (Amended) The chimeric oligonucleotide of claim 3, wherein said block of DNA [block] residues comprises at least 5 contiguous nucleotides.

6. (Amended) [The] A chimeric oligonucleotide [of claim 5, wherein said plant nucleotide sequence is a nucleotide sequence that encodes] comprising at least a first block of RNA residues and a second block of RNA residues, wherein said first and said second blocks of RNA residues are homologous to a plant herbicide resistance gene and flank a block of DNA residues, said oligonucleotide being capable of folding to form a duplex oligonucleotide.

11. (Amended) The chimeric oligonucleotide of claim 6, wherein said chimeric oligonucleotide is selected from [those set forth in Figures 1 through 13 (SEQ ID NOS: 1-13)] the group consisting of SEQ ID NO: 1, 2, 3, 4, 5, 6, 7, 8, 9, and 10.